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# ASCIDIANS COLLECTED AROUND THE GALAPAGOS ISLANDS USING THE *JOHNSON-SEA-LINK* RESEARCH SUBMERSIBLE

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*Abstract.*—Ascidians were collected for the first time by a manned submersible on ocean-bottoms between 300 and 800 meters around the Galapagos Islands. Solitary forms dominate; among the eight species collected, five are new ones: *Polyclinum johnsoni, Ciona pomponiae, Situla rineharti, Ascidia fusca,* and *Styela psoliformis.* They belong to surprisingly diverse families and several have deep-sea characteristics.

The fauna of the eastern part of the tropical Pacific Ocean is poorly known, especially the ascidians. Tokioka (1972) reported on a small collection of ascidians from the coast of Costa Rica, and recently Millar (1988) identified some specimens from the Galapagos and Ecuador. Nothing else is reported about ascidians between Baja California and Chile.

In November 1986, an expedition to the Galapagos Islands collected deep-water organisms for pharmacological studies. Over 600 animals were collected by the R/V *Seward Johnson* and *Johnson-Sea-Link-1* manned submersible (Harbor Branch Oceanographic Institution, Fort Pierce, FL) using a manipulator arm equipped with a claw, a suction tube, and a grab sampler. Among this collection there are eight species of ascidians, five of which are new species.

The material is particularly interesting because the specimens were collected undamaged on rocky bottoms, where a dredge or any blunt device monitored from a surface boat cannot operate. The animals are unusually large compared to the size of the ascidians collected in shallow water by SCUBA during the same cruise and relative to the deep-water ascidians on soft bottoms elsewhere.

Observation from the submersible during several dives suggests that ascidians are not evenly distributed on the bottom, but live in patches in restricted areas. They grow attached to vertical walls, in small caves, or on large cobbles; they are directly in contact with loose sediment.

The solitary forms seem more abundant in deep water than colonial ones, a situation opposite to that in shallow water areas outside of the Galapagos Islands. This may be due to the collecting method: solitary ascidians, especially large ones, are easily seen from the submersible and can be collected, but colonial ascidians normally live in crusts on the undersides of rocks or pebbles, or in deep crevices, and so escape notice. The colonies reported here were attached to cobbles.

Of the eight species collected, two are colonial. They both belong to the family Polyclinidae (order Aplousobranchiata). One is a new species of the genus *Polyclinum,* named *P. johnsoni* after the submersible and ship. The other species is probably *Aplidium californicum* (Ritter & Forsyth 1917), one of the most common Polyclinidae on the shore nearby, but the samples are too small and not fully enough developed to verify their identity. The other ascidians belong to surprisingly diverse families: one Cionidae, two Ascidiidae, and one Octacnemidae among the order Phlebobranchiata; and one Styelidae and one Pyuridae in the Stolido-

branchiata. Several of these solitary ascidians have deep-sea characteristics: *Ciona pomponiae,* n. sp. has a particularly solid, noncontractile tunic, and a very small gut compared to the branchial sac; *Situla rineharti,* n. sp. belongs to a typical bathyal family (Octacnemidae); *Styela psoliformis,* n. sp. has the same characteristics as other deep-water species of that genus.

The presence of a deep-water ascidian fauna around the Galapagos, different from the shore species collected by SCUBA, is not surprising. The bottom temperature at 500–700 m was between 7°C and 5.5°C. The water near the bottom was rich in suspended particles, but it is not known whether the particles were plankton except for the numerous crustacean larvae observed.

Type specimens are deposited at the Smithsonian Institution, National Museum of Natural History (USNM). Sorted samples received a Sea Pharm number (SP), indicated here for each specimen.

## Polyclinidae

### *Polyclinum johnsoni,* new species

Fig. 1A, B, C, D

*Holotype.*—USNM 18249; SP 25-XI-86-3-17.

*Description.*—Two colonies were collected close to Punta Espinosa, Fernandina Island, at 306 m depth, attached to a sponge underneath a large piece of rock. The colony retained as a taxonomic voucher is a cushion 2.5 cm in diameter. Color in life pale yellow with translucent tunic through which yellow zooids were clearly visible. Branchial sac and internal papillae obscured by an orange-yellow pigment. Zooids situated on either side of cloacal canals that converge towards the center of the colony where colony's sole common cloacal aperture opens.

The zooids have a large size range. Thorax large compared to the abdomen and the post-abdomen (Fig. 1A). Oral siphon star-shaped with six triangular pointed lobes above a strong sphincter that closes the siphon into a narrowed tube. Oral tentacles vary in number with the zooids and are inserted in at least three circles, with three or four orders of length, the largest being dorsal. At least 40 oral tentacles can be counted in the largest zooids. The peripharyngeal band lined with two high laminae and not curved at the level of the dorsal tubercle. Cloacal aperture close to the oral siphon and provided with a sphincter which may make the aperture into a tube when contracted. Dorsal lappet (Fig. 1A) well developed and inserted on a large base; not wide but its length, which varies according to the location of the zooid in the colony, can reach at least the length of the thorax; has two longitudinal muscular bands (Fig. 1A). There is a rounded button under the cloacal aperture, as is usual in the genus. Thoracic musculature is weak; longitudinal fibers from the oral siphon stop very quickly and do not pass the level of the first row of stigmata; there is an average of eight muscles on each side.

Branchial sac, in both the live and preserved animals, visible by transparency; the transverse vessels underlain by muscular fibers and bear high papillae (Fig. 1B) containing an orange-yellow opaque pigment. The number of rows of stigmata is variable according to size of zooids, and divisions of stigmatal rows commonly seen. In the colony studied there is a maximum of 16–17 rows of stigmata. In this example the fourth row is divided as follows: 25 stigmata on the right and 23 on the left side with 16 papillae on the right and 15 on the left on the fourth transverse vessel. Branchial sac not perforated along the endostyle or under the rapheal papillae. Dorsal languets short and wide, only slightly displaced to the left side. The whole branchial sac has a rectangular shape with a right angle at the base of the endostyle (Fig. 1A).

Abdomen slightly twisted; esophagus short; the stomach placed anteriorly is suddenly cut in its pyloric part (Fig. 1C). There is a ring-like post-stomach; intestine not

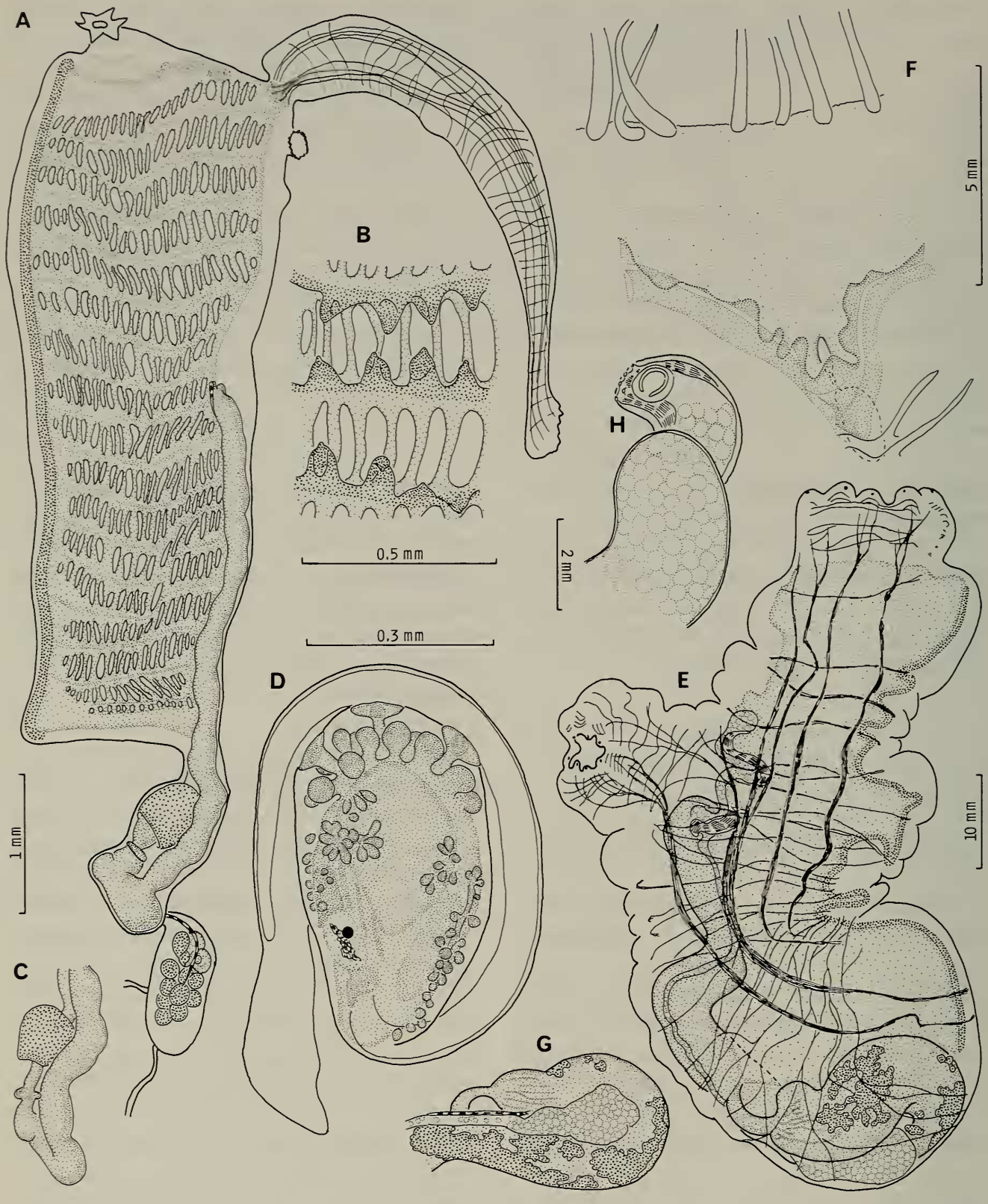


Fig. 1. *Polyclinum johnsoni*: A, Zooid; B, Detail of the branchial sac; C, Digestive tract; D, Larva. *Ciona pomponiae*: E, Animal without tunic, right side; F, Neural region; G, Gonads on the digestive loop; H, Extremities of gonoducts.

further differentiated into regions (Fig. 1C). Anus opens at the level of the 7th or 8th row of stigmata and has two lips.

Post-abdomen egg-shaped, linked to the abdomen by a thin peduncle (Fig. 1A); ovary is anterior followed by a cluster of testes. The heart is terminal. One or two vascular appendices extend from the post-abdomen into the test.

Larvae (Fig. 1D) incubated in the cloacal cavity. The zooids contain 1–3 larvae. The trunks measure 550 μm. They have 3 adhesive papillae, 7 pairs of thick epidermal papillae arranged on a line on each side of the body except for one pair which is lightly displaced inside, and there are two areas of epidermal vesicles on each side, one dorsal and the other ventral (Fig. 1D).

*Remarks.*—This species differs from all other *Polyclinum* with more than 15 stigmata rows in having higher papillae on the transverse bars, a large number of stigmata per row, and a peculiar distribution of the pigments in the colony. The genus *Polyclinum* is very homogeneous and the anatomical differences between the species are slight, so it is not surprising that deep-water species do not differ substantially from shallow-water species.

There is only one *Polyclinum* species known to live deeper than *P. johnsoni,* n. sp.; *P. neptunium* Hartmeyer, 1912, collected at 106 m and 318 m off the Cape of Good Hope.

The species is named after the submersible from which it was collected.

## Cionidae

### *Ciona pomponiae,* new species

Figs. 1E, F, G, H, 2A, B

*Holotype.*—USNM 18247, SP 21-XI-86-2-1.

*Description.*—The unique specimen measures 15 cm in length; attached by the left posterior side; oral siphon terminal; cloacal aperture located ⅓ the way down the dorsal side (Fig. 2A). Both siphons sessile. Test soft, gelatinous, colorless in life and free of foreign particles; is at least 5 mm thick and does not appear to consist of several layers as in *Ciona intestinalis.* Body contracted inside tunic and no longer attached to it, even at the level of the siphons; size of body reduced to 6.9 cm. Tunic much less retractile than in other *Ciona* species.

Musculature on each side (Fig. 1E) forms four muscular ribbons initiated from the oral siphon and two ribbons from the cloacal siphon. The two most dorsal muscles of the oral siphon and the one most ventral muscle from the cloacal siphon gather to form one ribbon in the posterior part of the body. The two ventral muscular bands issuing from the oral siphon become narrow before reaching the posterior part of the body and two muscles reach the bottom of the branchial sac (Fig. 1E). There is a network of transverse fibers. This small number of muscular bands is peculiar to this species.

There are few oral tentacles (16 large ones and several very small) placed without order on a crest. The length of some tentacles allows them to protrude through the oral aperture. The peripharyngeal band consists of two unequal laminae; the anterior thin and high, the posterior low and thick. There is a marked dorsal V in which the neural duct opens as a simple slit (Fig. 1F). Neural gland close to the nerve ganglion. Dorsal languets have an increased length posteriorly, measuring 6 mm in the anterior ⅓ and 18 mm near the esophagus.

Branchial sac (Fig. 2B) thin, the trabeculae connecting it to the body wall long. Transverse vessels of several orders regularly distributed; those of the first order wide and protrude into the branchial cavity, those of the third or fourth orders are thin laminae protruding into the branchial cavity. They bear large papillae that are flattened longitudinally and form hooks above the longitudinal bars. Longitudinal bars thin; stigmata elongated, generally numbering six to

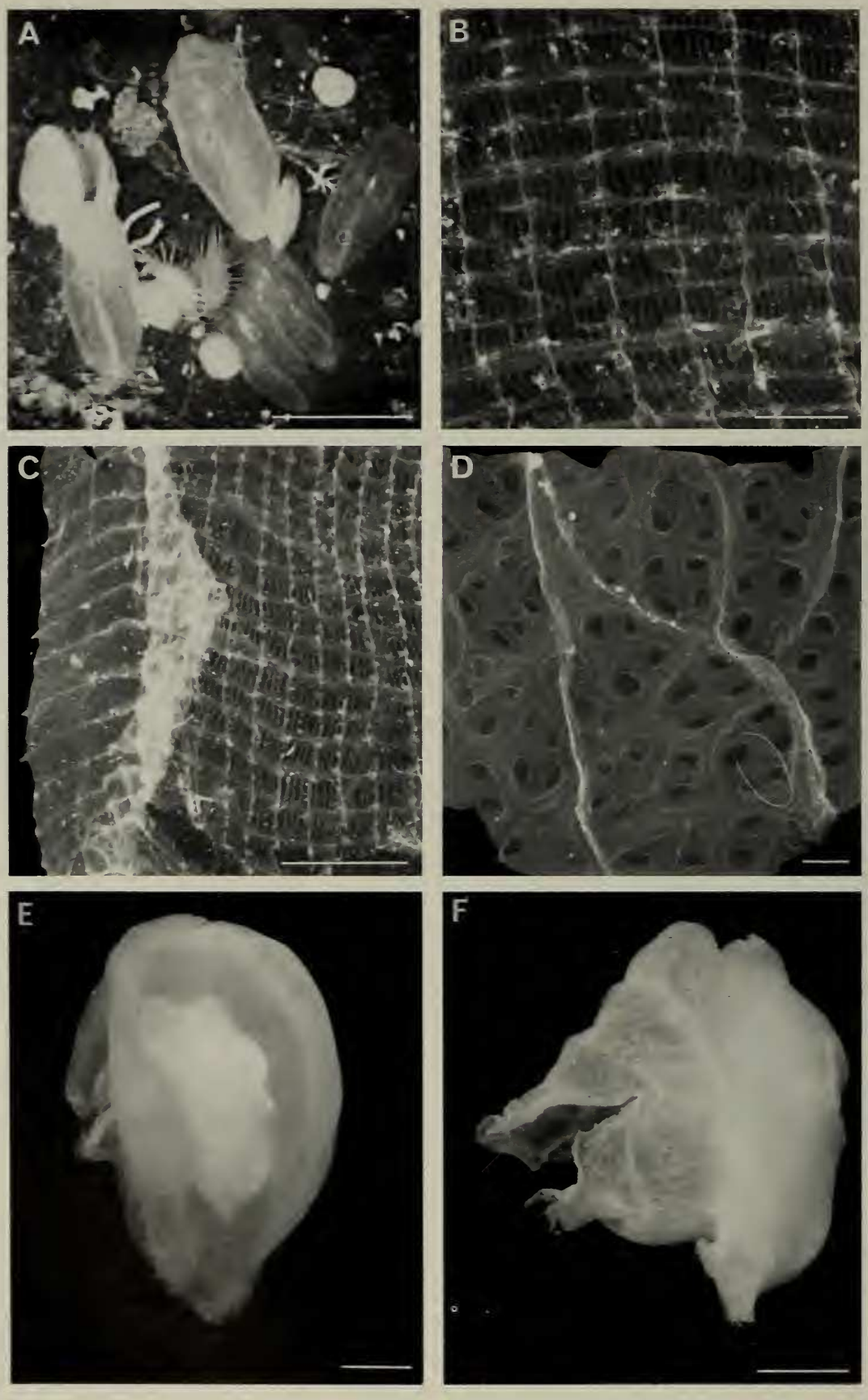


Fig. 2. *Ciona pomponiae*: A, Group of in situ individuals (from a submersible videotape); B, Part of branchial sac. C: *Ascidia clementea* part of the branchial sac with dorsal lamina. *Situla rineharti*: D, Part of the branchial sac; E, Habitus left side; F, The same without tunic. (Scales: A = 10 cm; B, C, D = 1 mm; E, F = 1 cm.)

eight in a mesh; most crossed by a high parastigmatic vessel bearing papillae (Fig. 2B); often divided under this vessel. If the division is complete, the stigmata on each side of the vessel are shorter than the others, and a very thin new parastigmatic vessel without papillae can occur above them.

Digestive tract clearly situated under the branchial sac; enlarged stomach has internal plications visible by transparency; most posterior fold prolonged and ends in a small ampulla at its junction with the intestine; intestinal wall thin and gut contents can be seen through it. Rectum elongated and ends in a non-lobated narrow anus; posterior intestine encircled by a well developed pyloric gland.

The pear-shaped ovary is located under the intestinal loop (Fig. 1E, G). Testis diffuse, consisting of numerous acini which coat the intestinal wall; ducts join to form a common sperm duct at the level of the anterior part of the ovary; i.e., oviduct and common sperm duct originate at the same point. Gonoducts contain large numbers of eggs (400 $\mu$m in diameter) and spermatozoa, and pass beyond the anus; oviduct opens in a flat horseshoe-shaped papillae (Fig. 1H). Male papilla has several small apertures; there is no pigment spot at this level.

*Remarks.*—This species is very uncommon for a *Ciona.* Although very few species occur in shallow waters, the genus is better differentiated deeper on the continental slope. In the northeast Atlantic is found *Ciona gelatinosa* Bonnevie, 1896 and *Ciona imperfecta* Monniot & Monniot, 1977, and in the northeast Pacific *Ciona mollis* Ritter, 1907.

*Ciona mollis,* known from California at 2011 m depth off San Nicolas Island, differs from the new species in several important characters: *C. mollis*'s size is smaller (3 cm maximum) and its tunic is very soft. There are 6 muscle bands on each side of the body, all of them reaching the posterior part where they fuse. There is circular musculature only on the siphons. The oral tentacles are very numerous, more than 200, while the present specimen, much larger, has only 16. This difference is perhaps not as significant as the reduction of the number of oral tentacles even with increased size in *Ascidia translucida* according to Millar (1960) and in *Cnemidocarpa verrucosa* according to Monniot (1978). In *C. mollis,* furthermore, the digestive tract does not have the same shape as in this specimen, *C. mollis*'s intestine curves immediately after the stomach, and its gonoducts do not open near the anus. *Ciona mollis* seems closer to the Atlantic species *C. gelatinosa* Bonnevie 1896, redescribed by C. Monniot (1970), in the musculature (7 bands), the shape of the gut, and the location of the genital papillae.

The species is named after Shirley Pomponi, co-chief scientist of the cruise.

Octacnemidae

*Situla rineharti,* new species

Figs. 2D, E, F, 3A, B, C, D

*Holotype.*—USNM 18244; paratype USNM 18245; SP 14-XI-86-1-2 and 18-XI-86-2-11.

*Description.*—Among several samples collected, two specimens have been examined: one was designated as the holotype, collected north of Floreana (Santa Maria Island) at 790 m depth, and another (paratype) collected south of Genovesa (Tower Island) at 695 m depth. All specimens collected are approximately the same size, almost spherical, with a cartilaginous white tunic that is translucent more than transparent (Fig. 2E). Holotype ovoid, attached by the small ventral end (7 × 5 × 5.2 cm). The part of the tunic attached to the substratum torn but represents a narrow area of attachment; in other specimens there is no particular differentiation of the attachment area. Oral aperture a gaping transverse slit, made of two large lips located at half the height of the body (Figs. 2F, 3A). Dorsal lip protrudes more than ventral one. Cloacal

aperture not obvious; it is a simple hole at the upper part of the body slightly displaced to the dorsal side (Fig. 3A). Tunic on the two lips of the oral siphon is thin, cartilaginous, and glossy, radially wrinkled, and different from the thick test (at least 3 mm) around the rest of the body. Border between the two kinds of tunic marked by a groove at the limit of the lips.

Removed from the tunic, the body appears composed of three parts: the oral siphon formed of two well developed lips, a branchial cavity widely spread separated from the oral siphon by a ring of tentacles, and a "nucleus" containing the gut and gonads.

The musculature is complex (Fig. 3A, B). The muscles of the oral siphon, independent of the body musculature, include radial fibers regularly distributed on both lips; these end at the circle of oral tentacles and the peripharyngeal groove. The oral siphon's circular fibers are also regularly placed, spread over both lips and gathered at the lip corners. At each of those corners is a fan of fibers belonging to the radial musculature, connecting the corners to the large muscle located at the level of the ring of oral tentacles.

Musculature of the cloacal siphon consists of radial and circular fibers, weakly developed and anastomosed. It is limited to the immediate vicinity of the siphon.

A large, strong ribbon of muscle fibers encircles the body above the ring of oral tentacles, and has a horseshoe shape opened on each side of the endostyle at the level where the body attaches to the substratum (Fig. 3A). A muscular string binds the ribbon's two free extremities. There are also two areas of transverse muscles dorsally, and ventrally two bundles more or less parallel to the endostyle, connected to the "string" linking the posterior extremities of the large horseshoe muscle ribbon.

Oral tentacles (Fig. 3C, D) situated on a rim located posterior to the horseshoe muscle-band. Dorsally the rim is low; its height increases ventrally where it is lined by a flat muscular band. Tentacles have a unique leaf-shaped structure, narrowed at their insertion on the rim. On the anterior side of each tentacle is an ampulla which stains very deeply with hemalum. The tentacles are of two regularly alternating sizes (Fig. 3C).

The peripharyngeal groove is far from the ring of tentacles in the dorsal half of the body but comes close to it in the ventral part. Dorsally, a series of irregular crests separate the peripharyngeal groove and the tentacles and they may be considered as the anterior rim of the peripharyngeal groove. Dorsally the peripharyngeal groove is deeply incurved (Fig. 3D) to reach the dorsal tubercle. The latter is rather far from the neural ganglion; its aperture is a simple hole. The neural gland is located on the left posterior side of the neural ganglion.

There is no differentiated dorsal lamina. The restricted space between the dorsal tubercle and the opening of the esophagus is marked by transverse elevations continuous with more or less regular internal transverse crests on the branchial sac; these elevations can be considered as transverse vessels. There is a short endostyle originating near the oral tentacles and ending far from the entrance of the esophagus (Fig. 3A, B). At its posterior extremity, on the left, is a retropharyngeal groove which initially consists of a single rim but divides in two wide and flat ridges.

The entire surface placed between the peripharyngeal band and the branchial stigmata is covered with small papillae. Branchial cavity cup-shaped with the bottom close to the digestive tract. The perforations, limited to two small areas in the flat bottom of the cup on each side of the retropharyngeal band, lie in thick tissue. The perforations are oval (Fig. 2D) and generally grouped in pairs; they are not ciliated, and are larger than in the other known species of Octacnemidae.

The digestive tract forms a simple loop (Fig. 3B); esophagus opens by a simple hole

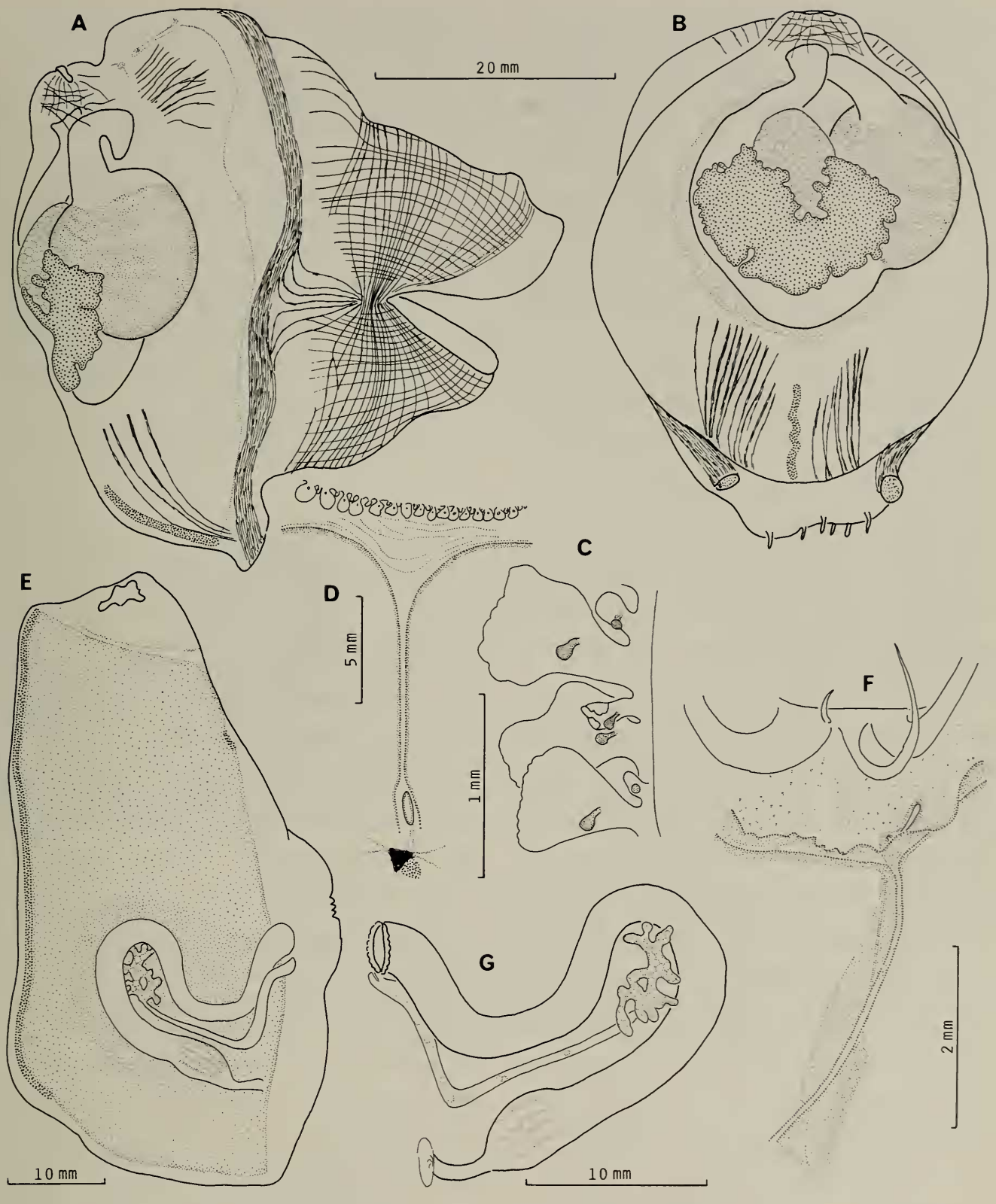


Fig. 3. *Situla rineharti*: A, Right side of the animal without tunic; B, Postero-dorsal side; C, Oral tentacles; D, Peripharyngeal groove indentation,tentacles and neural ganglion. *Ascidia clementea*: E, Left side of the body, tunic removed; F, Dorsal part of the tentacle ring and peripharyngeal groove; G, Intestinal loop with ovary and oviduct.

in the branchial sac; stomach is wide and has a glandular wall which appears somewhat wrinkled but has no internal plications (Fig. 3A, B); its external oblique ridges may be due to contraction. The intestine is thin and transparent, ending in a plain-edged anus placed beside the esophageal entrance. The digestive tract and gonads are embedded in a thick mesenchyme. The retropharyngeal groove lies beside the middle of the gut loop and indicates that the stomach is on the right side.

The gonads (Fig. 3A, B) consist of a massive and protrusive ovary located between the two branches of the digestive loop; testis is composed of numerous acini; it is flat and covers the ventral part of the ovary and each side of the intestine. The oviduct opens on the medial side of the intestine a short distance before the anus. The spermduct, full of spermatozoa, opens next to the oviducal aperture on a small papilla. The heart lies alongside the intestine.

*Remarks.*—Octacnemidae having an oral aperture with two large lips belong to four genera: *Dicopia* Sluiter, 1905; *Megalodicopia* Oka, 1918; *Situla* Vinogradova, 1969; and *Cibacapsa* Monniot & Monniot, 1983. *Cibacapsa* is characterized by the total absence of a branchial sac. The distinctions between the other genera are less obvious. The difference between *Dicopia* and *Megalodicopia* is the presence in the latter of a peduncle, which in our opinion is not a difference of much importance. *Dicopia* differs from *Situla* by the presence in *Dicopia* of a true branchial cavity with a conical perforated area and oral tentacles forming a ring of a smaller diameter than that of the branchial cavity. In *Situla* the branchial cavity opens widely and the perforated area is in a flat disk around the digestive tract. The Galapagos species is closer to *Situla.* The main differences between the species of *Dicopia* and *Situla* are summarized in Table 1. The new species is the only one having a large imperforate space between the peripharyngeal groove and the branchial tissue, and its oral tentacles have a very original structure.

Ritter (1907) has incompletely described a species of Octacnemidae collected off San Diego, California: *Benthascidia michaelseni.* His account suggests he did not understand the anatomy of the animal very well. The two specimens of that species preserved at the Smithsonian Institution are dissected and their organs isolated, and so it is impossible to reconstitute their original disposition in the animal. In any case Ritter's material cannot be of the same species as the new Galapagos one.

The species is named after Kenneth Rinehart, co-chief scientist of the cruise.

Ascidiidae

*Ascidia clementea,* Ritter, 1907

Figs. 2C, 3E, F, G

SP 23-XI-86-3-27.

*Description.*—One specimen was collected north of Isabella Island at 335 m depth. The ovoid body is attached to the substratum by the posterior part of the left side. The maximum length of the animal is 6.8 cm. Oral aperture terminal; cloacal aperture located at the anterior ⅓, the distance between the siphons being 2.9 cm. The tunic in life is yellowish, covered with small cylindro-conical papillae. Papillae and grooves of the tunic are pigmented with ochre; both apertures are 6-lobed. The test is partially covered with epibionts: foraminifera, polychaete tubes, bryozoans and molluscan egg-capsules. One of the cheilostome bryozoans consists of thecal rows, each zooid having a long ventral extension penetrating deeply into the ascidian's tunic; the bryozoan colony is raised above the tunic surface by these extensions. Thickness of the tunic averages 2 mm; its internal side is soft and translucent.

Body wall thin and transparent; the musculature forms 2 bands of transverse parallel fibers—a well developed dorsal band, and a smaller ventral band—and longitudinal

Table 1.—Comparisons between the species in *Situla, Dicopia* and *Megalodicopia* genera.

| Name | Distribution | Body shape | Tentacles | Sensory papillae | Peripharyngeal groove | Peritubercular area | Branchial sac |
|---|---|---|---|---|---|---|---|
| *Dicopia fimbriata* Sluiter, 1905 | Indonesia | hairy sphere, 2 large lips | not seen | not seen | close to tentacles and branchial sac | short | cone shaped |
| *Dicopia japonica* Oka, 1913 | Japan | Sphere with papillae, 2 large lips | absent ! | not seen | close to tentacles and branchial sac | short | cone shaped |
| *Megalodicopia hians* Oka, 1918 | Japan | peduncle, short lips, smooth tunic | on a scalloped line | between tentacles and peripharyngeal groove | close to tentacles and branchial sac | short | cone shaped |
| Tokioka, 1953 | Japan | | | | | | |
| Kott, 1969 | Weddell sea | large lips, hairy tunic | | | | | |
| *Situla pelliculosa* Vinogradova, 1969 | Kurile trench | short peduncle, large lips, thin smooth tunic | on a raised ring | between tentacles and peripharyngeal groove | close to branchial sac | short | flat fully perforated ring |
| *Dicopia antirrhinum* Monniot C., 1972 | Europe | peduncled or sessile, large lips, thin hairy tunic | on a raised ring | anterior to tentacles | close to tentacles and branchial sac | short | cone shaped |
| *Situla lanosa* Monniot C., & F., 1973 | Europe Mayotte | short peduncle, large lips, thin hairy tunic | on a line with 2 inflexions | between tentacles and peripharyngeal groove | close to branchial sac | short | flat ring, laterally perforated |
| *Situla multitentaculata* Vinogradova, 1975 | Scottia sea | peduncle, no lips, thin smooth tunic | oval line | not described | dorsally far from branchial sac but near ventrally | long | flat fully perforated ring |
| *Situla rebainsi* Vinogradova, 1975 | Scottia sea | peduncle, large lips, smooth tunic | oval line | between tentacles and peripharyngeal groove | far from tentacles, close to branchial sac | short | flat fully perforated ring |
| *Situla macdonaldi* Monniot C., & F., 1977 | South Indian Ocean (MacDonald Island) | short peduncle, large lips, thin hairy tunic | scalloped line with inflexions | between tentacles and peripharyngeal groove | close to branchial sac, far from tentacles | short | flat fully perforated ring |
| *Situal rineharti,* n. sp. | Galapagos | sphere, short lips, smooth thick tunic | raised circle | between peripharyngeal groove and branchial sac | far from tentacles, far from branchial sac dorsally | long | 2 flat areas at the bottom of a cup |

muscle fibers extend from the oral siphon. On the right side all the muscles spread and intermingle to make a loose network. On the left side the longitudinal fibers extend only a short way from the oral siphon, and most of the left body wall is devoid of musculature.

Seventy oral tentacles arise from an elevated rim that has a muscular ring. The tentacles are of at least four orders of size, regularly alternated although the smallest ones are not developed everywhere. Peripharyngeal groove made of two unequal laminae, the anterior one being larger, is close to the tentacles, and the space between is covered with thin papillae (Fig. 3F). It does not make a deep 'V' dorsally; the dorsal tubercle is low, J-shaped. Neural ganglion distant from the dorsal tubercle; neural duct is twice the length of the neural ganglion. Rapheal band consists of two laminae which measure five times the length of the dorsal tubercle. Posteriorly, its edge is higher and carries teeth that coincide with the transverse vessels (Fig. 2C). After passing around the esophageal entrance it becomes irregular. On the right side close to the esophagus the transverse vessels end without differentiation.

Branchial sac thin and flat (Fig. 2C). At the level of the top of the intestinal loop there are 100 longitudinal vessels. Main papillae are located at the junctions of the longitudinal and transverse vessels, and almost everywhere in the branchial sac there are smaller intermediate papillae, which are not linked to the presence of parastigmatic vessels. These vessels exist only where there is duplication of a stigmata row. There are four to six stigmata in a mesh, except on the right side, where the meshes located near the esophagus have more than 20 stigmata.

The gut forms an open loop with an accentuated secondary curve (Fig. 3E). The stomach appears plicated externally (Fig. 3E, G). The intestine is deeply embedded in a mass of large renal vesicles which spread over the body wall. The anus is attached to the body wall and has two slightly scalloped lips. The specimen was not sexually mature. The ovary, a network of tubes inside the intestinal loop, does not spread out onto the intestine (Fig. 3G). We have not found the testicular acini that Ritter (1907) shows against the stomach.

*Remarks.*—*A. clementea* has not been reported since its description in 1907 based on eight specimens: one collected by the *Albatross* near San Clemente Island at 654–704 fathoms (station 4405), and seven specimens at 21.8 miles to the south of San Nicolas Island, 1100 fathoms (station 4425). There are a few small differences from Ritter's description: he does not mention the tunic papillae and counts eight lobes at the oral siphon instead of six. His figure 33 shows more than 75 small oral tentacles all of equal size. He describes "deep narrow plications" of the branchial sac, but his fig. 34 does not show this. In his discussion, he recognizes that these plications do not always exist. All the other characters of our specimen are identical to his description.

*Ascidia fusca,* new species
Fig. 4A, B, C, D

*Holotype.*—USNM 18248; SP 20-XI-86-1-25.

*Description.*—The sole specimen was taken at 375 m depth southeast of San Salvador (James) Island. Maximum length 5.6 cm; it was lying on its left side. Apertures distant from each other (4.6 cm); have an undetermined number of external large soft swellings. Tunic dark brown in life (thus the species name *fusca*) and its surface wrinkled. Some foraminifera and sedimentary particles adhere to the settlement surface; elsewhere on the tunic are small epibionts, mostly bryozoans. The test is soft and its internal layer is colorless.

Musculature strong and lies entirely on the right side; is interrupted before it reaches the endostyle and dorsal lamina; comprises one ventral and one dorsal field, which intermix in the middle of the right side. The fields' fibers are close together and nearly

joined; siphonal musculature weak. On the right side of the oral aperture an array of about 10 bands blends into both transverse fields and disappears in the first ⅕ of the body.

Oral tentacles arise from posterior part of a velum linked by a muscular ring; there are about 40 large ones and as many very small ones alternating and irregularly developed; bases crooked; peripharyngeal groove made of two unequal laminae: anterior high and thin and the posterior forming a flat ribbon. There is a large space between the tentacle ring and the peripharyngeal groove which is indented into a deep dorsal V (Fig. 4B). Dorsal tubercle is urn-shaped, opening with a simple aperture (Fig. 4B). Neural ganglion far from the dorsal tubercle at a distance of 3 times its length (0.6 cm in the type specimen). Dorsal lamina has 2 rims which extend to the posterior part of the neural ganglion and then form a single lamina that is thin and high. This carries digitiform papillae which prolong the transverse sinuses. The lamina becomes progressively lower and somewhat irregular as it nears the esophagus, where it forms a low, toothed crest. At this level on the right side, one of two transverse vessels ends in a large lanceolate papilla.

Branchial sac flat; at the level of the top of the intestinal loop there are 70 longitudinal vessels on the right and 55 on the left, the left side being much more narrow than the right side. Longitudinal vessels very thin and carry fingerlike main papillae where they intersect transverse vessels. There are neither secondary papillae nor parastigmatic vessels. The meshes are elongate and contain 2 to 3 stigmata.

Gut (Fig. 4A, C, D) small; stomach, clearly enlarged, has some plications only visible on its medial side. Intestine inserted inside the wall of the mantle. The 2 lips of the anus have many small lobes. In this specimen a translucent swelling is located close to the anus. It has no discernable internal structure and we do not know if it is a specific character or an artefact.

The ovary (Fig. 4C, D) consists of a mass of tubes projecting internally, with edges growing partly over the inside of the intestine. The testicular lobules lie on the 2 sides of the gut. The gonoducts open together next to the anus. The renal vesicles are small, almost invisible.

*Remarks.*—*Ascidia fusca* differs from *A. vermiformis* (Ritter 1913), another deep-water *Ascidia* in the East Pacific, by the former's external shape, the length of its cloacal siphon and the long posterior extension of its branchial sac. As in several of the rare *Ascidia* known from the continental and island slopes, *Ascidia fusca* has a relatively small gut compared to the branchial sac, an open primary gut loop, and a flat branchial sac.

The species name refers to the dark tunic of the animal.

Styelidae

*Styela psoliformis,* new species

Fig. 4E, F, G

*Holotype.*—USNM 18246, SP 16-XI-86-1.

*Description.*—The sole specimen was found in a cavity of a basaltic cobble near Espanola island at 567 m depth. The visible part of the still attached body is an oval disk, 14 × 11 mm, white, covered with low, round, plate-like papillae which have a vascular dilation full of blood granules. Around the disk the tunic is thinner and also has vascular ampullae; it strongly attaches the animal to the substratum. Both apertures protrude a distance of 6 mm, but they are located in depressions, so they do not extend beyond the body surface after contraction. Tunic of the central disk opaque and internally pearly. The rest of the body has a thinner tunic that is soft and transparent. Inside the tunic the body is flattened against the substrate. The dorsal part of the body wall is opaque and muscular, the ventral side thin and transparent without musculature. At the boundary between the dorsal and ventral side the mantle has a ring of

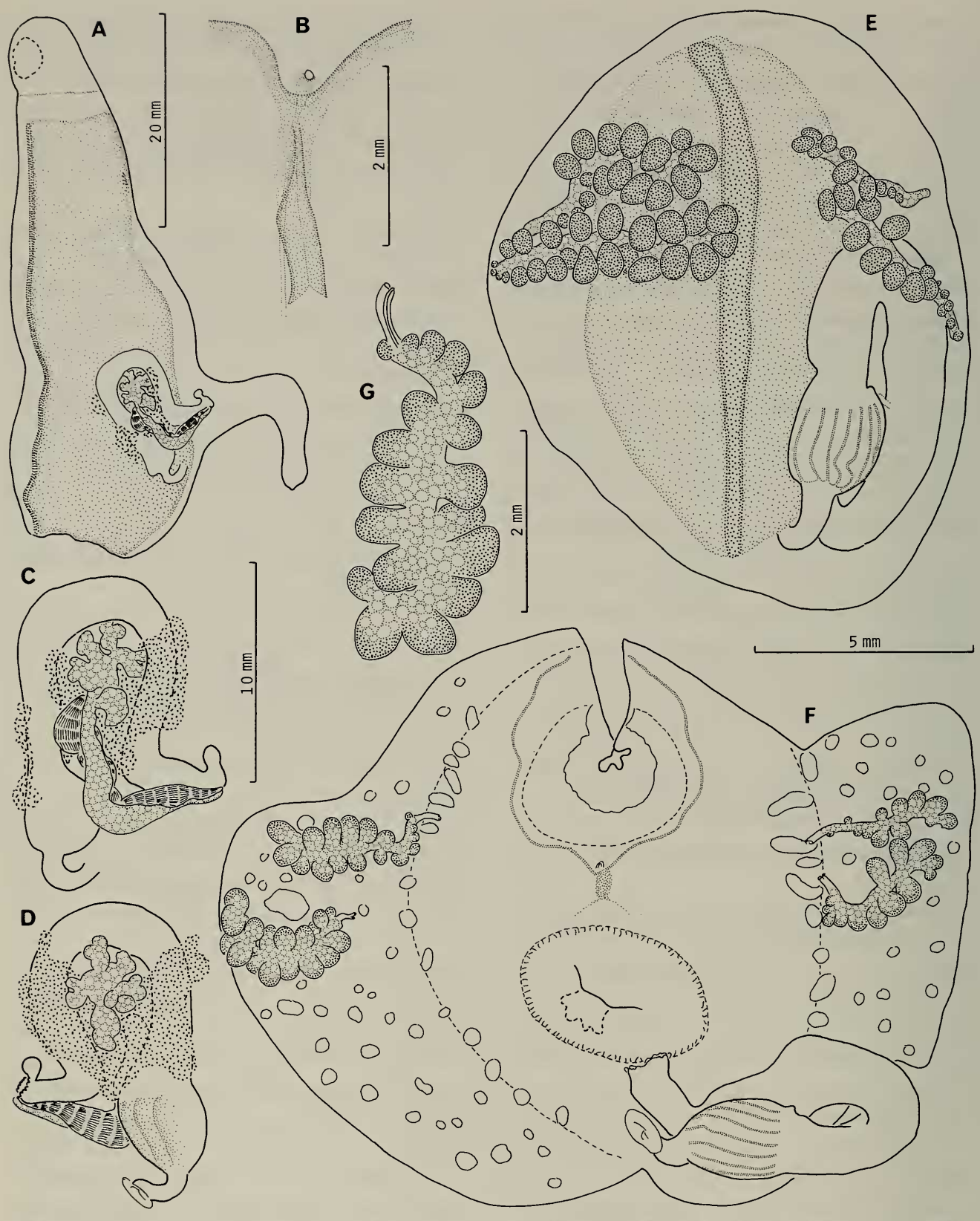


Fig. 4. *Ascidia fusca*: A, Left side of the body, tunic removed; B, Dorsal tubercle; C, External side of the gut and gonads; D, Internal side of the gut and gonads. *Styela psoliformis*: E, Ventral side of the body, tunic removed; F, Internal side of the body, branchial side removed; G, Gonad.

tissue rich in tan blood lacunae clearly seen by transparency.

About 30 oral tentacles are attached to a thin crest located posteriorly to a large oral velum. There are at least four different sizes which regularly alternate. The oral tentacles are wide and transversally flattened. The peripharyngeal groove has 2 equally pro-

truding rims. It is far from the tentacles on the ventral part, nearer on the sides, and dorsally makes a little but distinct V; it is only curved around branchial folds number 1 and 2. The opening of the dorsal tubercle has the shape of a C opened on the left. The neural ganglion is in contact with it. The dorsal lamina is high and smooth.

Branchial sac has 4 folds on each side in the anterior part only; fold number 4 disappears in the posterior ⅔ of the branchial sac. Longitudinal vessels very numerous and difficult to count; there are as many as 15 vessels between the folds and more than 20 on folds 1 and 3. Fold 4 has more than 15 longitudinal vessels anteriorly. The folds are high and the longitudinal vessels close to each other, with only one or two elongated stigmata per mesh. Parastigmatic vessels are generally present.

Gut a narrow loop (Fig. 4E, F). Stomach and intestine weakly linked to the ventral side of the body wall but rectum strongly united to dorsal side. The globular stomach has 14 obvious plications. A very small caecum is located near the gastric aperture of the pyloric gland. Intestine long; the short rectum opens by a wide, lobed anus.

There are two gonads on each side (Fig. 4E, F), weakly linked to the body wall; each is made of a double series of testes (Fig. 4G). The central ovary of each gonad spreads onto the internal part of these lobes, which makes this structure intermediate between the genera *Styela* and *Cnemidocarpa*. The gonoducts open together at the margin of the dorsal disk of the body wall.

Endocarps are evenly distributed on the ventral side of the body (Fig. 4F). They are very numerous and protrude along the ring of tissue rich in tan blood lacunae which marks the boundary between dorsal and ventral halves of the body. The dorsal part is devoid of endocarps. The cloacal aperture is circled by thin cloacal tentacles arising from a crest (Fig. 4F).

*Remarks.*—This species has several characters which are similar to *Styela milleri* Ritter, 1907, collected frequently from 900 to 4000 m depth along the Pacific coast from California to Chile and also recorded in the south Atlantic and the Indian Oceans. The oral tentacles, dorsal tubercle, dorsal lamina, branchial sac, and gut are similar in both species. In contrast, *S. milleri* has only one gonad on each side and they are different in structure from those of *S. psoliformis*. The new species has the same appearance as *S. similis* C. Monniot, 1969 from the continental slope of Europe and West Africa, and these species have many anatomical characters in common due to their similar body shape. *S. similis* also has only one gonad on each side, but its structure differs.

The gonad of *S. psoliformis* is morphologically intermediate between typical *Styela* gonads and the very peculiar gonad of *Cnemidocarpa pfefferi* (Michaelsen, 1904) shown in Millar 1960; that large (5 cm), round species lives on the slope of South Georgia and South Shetland Islands. It has two gonads on each side, each consisting of a lobed ovary with each lobe encircled by testes. The structure of the gonad of *S. psoliformis* resembles that of species of *Pyura*, but this is only a convergence: all other characteristics of the present species are typical of the Styelidae family.

The animal looks like a psolid holothurian after which it is named.

Pyuridae

*Halocynthia hispida* Herdman, 1881

Fig. 5

Synonymy: see Kott, 1985:342, fig. 169, pl. 7F, Millar (1988).

SP 24-XI-86-3-9 and 27-XI-86-1-9.

*Description.*—This species has two different types of morphology on the Galapagos Islands: a hard form with a thick tunic collected in shallow water by SCUBA and a larger form with a soft hairy tunic collected from deep water by the submersible. The two forms also differ in internal structure, and only a very careful analysis of all characters has convinced us that they belong to the same species. A reduction of the organs

linked to increased depth and decreased food supply has been described in ascidians (Monniot & Monniot 1978), but the case of *H. hispida* in the Galapagos is extreme.

The shallow-water form was collected at Bainbridge Rocks, east of San Salvador Island, at a depth of 17 m. It was firmly attached by its ventral side; both apertures are erect, at almost the same level. The specimen measures 10 × 7 × 5 cm, the apertures separated by 4 cm. The side facing the light is dark red in life. The tunic is wrinkled and covered with small smooth spines that become larger near the siphons, the largest grouped on tubercles. Around the apertures there are large branched spines 3 to 4 mm in length. The thickness of the cartilaginous tunic reaches 5 mm.

The body wall is 2–3 mm thick and fleshy. The muscles are well developed: the external circular ones make a continuous layer, the internal radial muscles are gathered together in strong bands. The oral tentacles are located behind a strong sphincter; they number about 20 in three orders and are only weakly pinnate. The peripharyngeal groove has 2 equal rims and shows a clear dorsal V. The dorsal tubercle is large, complex and protruding (Fig. 5D). The dorsal lamina has a double series of languets.

The middle level of the branchial sac has 10 high folds on each side. Anteriorly and posteriorly, additional ventral folds appear, so the maximum number of folds in this specimen is 13 on the right and 12 on the left side. The distance between 2 longitudinal vessels is the same on the folds as between them, so it is difficult to distinguish the base of a fold. In the ventral part of the branchial sac there are only 2 or 3 longitudinal vessels between folds, the meshes are larger there and they may have a dozen stigmata.

The gut (Fig. 5C) is voluminous and occupies most of the left side. The esophagus is long and the stomach is covered with a hepatic gland of two parts: the anterior lamellar and the posterior cauliflower-like. The intestine is attached to the body wall and has a smooth edged bilobed anus.

The gonads are very numerous (13 on the right and 10 on the left) and so massed and interpenetrated (Fig. 5C) that they can only be counted by their genital papillae. On the left side they cover the descending limb of the intestine. Each gonad consists of an ovary surrounded by lobes of testes.

There are numerous endocarps on the body wall, inside the gut loop and even protruding from the anterior part of the stomach (Fig. 5C).

The deep-water form is rounded; the largest specimen measures 7 × 5 × 4 cm with a distance of 4 cm between the apertures. The color in life is tan-brown with some amounts of red near the siphons. The entire tunic bears short branched spines with much larger ones on the siphons; between the branched spines there are smaller simple ones, often placed in rings. The test is thin (1 mm) and flexible. Internal organs are visible through the thin, transparent body wall. The external layer of circular muscles is thin. The internal radial muscles are gathered together in weak ribbons.

When the tunic is removed, the oral aperture has 4 low lobes with undulated edges. There is a small plain velum at the base of the oral siphon. The oral tentacles are not numerous (36) and occur in at least 4 orders. Between the largest tentacles on the tentacle crest there are numerous small digitiform ones. The largest tentacles measure up to 13 mm but only have about 20 primary branchings, which are themselves thin and poorly branched in turn. The peripharyngeal groove has 2 equal laminae; it is curved around each branchial fold and makes a large dorsal V. The dorsal tubercle is round and has a C-shaped aperture open on the anterior left side (Fig. 5A). The neural ganglion is elongated (15 mm), and thin. Nerves from it are clearly visible: 2 anterior and 2 posterior ones. The visceral nerve follows the dorsal lamina and is accompanied by a neat dorsal band; it originates on the right pos-

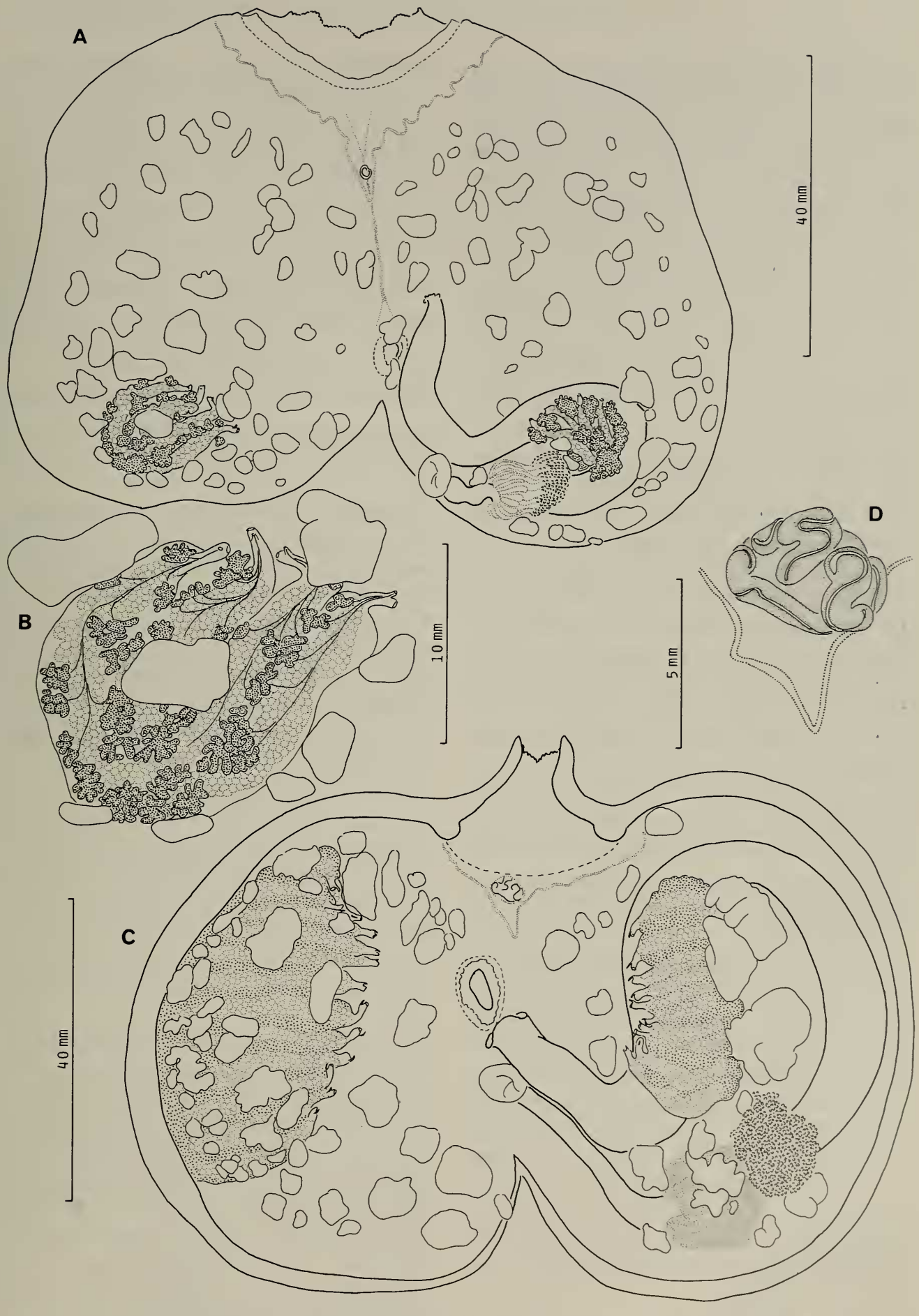


Fig. 5. *Halocynthia hispida*: A, Deep-water form, internal side of the body, branchial sac removed; B, Detail of the right gonad; C, Shallow-water form, internal side of the body; D, Dorsal tubercle of the shallow-water specimen.

terior side of the neural ganglion. In its anterior part the dorsal lamina consists of a double row of languets; the left line has true languets and is more developed than the right line, which is made of prolongations of the right transverse branchial vessels.

The branchial sac has 10 folds on each side. The branchial formula is:

R.E.2(8)2(13)2(15)3(17)2(18)2(20)2(22)2(21)3(20)3(16)2.DL

L.DL.2(17)2(21)2(22)3(22)2(21)2(19)3(15)2(13)2(11)1(10)O.R

The 10th (most ventral) fold on the left is only present in the anterior part of the branchial sac. The folds are very high and largely cover each other. The longitudinal vessels are raised, and there are only two or three between the folds. We counted 24 stigmata in a median mesh between two folds and 8 to 10 in meshes close to the base of the folds. It is only at the crest of the fold that the number of stigmata in a mesh go down to five or six. We have not observed spiral stigmata at the crests of folds. The stigmata are not very elongated; they are crossed by parastigmatic vessels only when a division begins in a transverse row. Between the folds the transverse vessels are inflated into ampullae attaining several millimeters, much like endocarps.

Gut short; a long esophagus leads to a stomach with a 2-part liver (Fig. 5A), the anterior one with parallel ridges and the posterior with papillae on crests. The two parts of the gland are separated by a narrowing, but they remain united. At the narrowing level there are parallel riges, each dividing in two in the anterior part (Fig. 5A). At the pyloric end of the stomach the hepatic gland stops abruptly. The intestine is short and translucent. Rectum long; attached along the dorsal lamina, and the anus has two lobed lips.

On each side there is a cluster of four parallel ovaries united at their proximal ends. Each cylindrical ovary is surrounded by ramified testes (Fig. 5B). Each ovary and its testes are wrapped in a common membrane. The testes give off long thin ducts which join at the atrial surface of the ovary to form a common spermduct. The common spermduct consists of canals coming from both sides of the ovary. The spermduct opens just behind the oviduct on a common protruding papilla. The four left ovaries originate inside the gut loop and open onto the intestine (Fig. 5A).

The entire body wall is provided with endocarps. The cloacal siphon has a short velum similar to the oral siphon. We have not seen cloacal tentacles.

*Remarks.*—This species is recorded from the Galapagos by Millar (1988) and the specimens he describes are intermediate between our two forms. If we examine point by point the differences between our shallow and deep populations, we find that they are almost all quantitative ones: thickness of the tunic and body wall, gonad number, size of the gut, and number of branchial folds. The difference in the number of oral tentacles has no taxonomic significance. The most important contrast is the reduction of the gut in deep-water specimens, and its position at the posterior part of the body. The gonads are also posteriorly displaced in deep-water animals.

The thinning of the tissues in deep-water animals is probably linked to decreased food supply. The species of *Halocynthia* have a highly evolved branchial sac which is very efficient in filtering phytoplankton, but the deep samples certainly have less food available. In fact, the branchial sac is the only organ not reduced; on the contrary, the number of stigmata per mesh increases with depth. The reduction in size of the gut is correlated with this nutritional phenomenon; it is a common characteristic for ascidians living on the slope such as in *Ascidia* (see below). A similar but less accentuated situation has been described for *Ascidia*

*challengeri* and *Corella eumyota* (Monniot & Monniot 1983) on the slope in the Antarctic Ocean and at Kerguelen.

The shifting of the gut and gonads posteriorly may be a consequence of the loss of tunic rigidity. In 1973 we reported that deep-sea ascidians show a tendency to equilibrate their visceral masses to keep their balance on the substratum. In other species of *Halocynthia* the disposition of the gonads on the right side seems linked to the tunic rigidity: *H. ritteri* from Japan has a hard tunic and gonads located in the middle of the right side, while *H. papillosa, H. aurantium* and *H. pyriformis,* with thin tunics, have gonads located posteriorly.

Except for two Arctic species, the tropical and temperate *Halocynthia* are found in very shallow water not deeper than 30 meters. Is the presence of deep specimens in the Galapagos exceptional? We cannot answer because the deep rocky bottoms have not been sampled enough, but this species represents the most extreme case of ecological dimorphism known in an ascidian.

## Conclusions

The bathyal ascidian fauna of the Galapagos does not provide any evidence in support of an archaic fauna. All species belong to genera and more advanced families which are well represented in other oceans and which show adaptations here to deep-sea life.

The origin of the fauna is unknown. Relatively few collections have been made along the South American Pacific coast; we cannot determine if the new species described here are endemic or if they have been carried north by the Humboldt Current. The Galapagos ascidian fauna differs greatly from the assemblage of Antarctic ascidians known from the south coast of Chile. Among the eight species recorded from the Galapagos, only three were previously known: *Aplidium californicum,* a tropical shallow-water shallow-water species probably at its depth limit here, *Halocynthia hispida,* widely distributed and known in shallow water from both the Galapagos and Australia; and *Ascidia clementea,* a north Pacific species. *Situla* and *Ciona* are genera which are well represented in bathyal areas in other parts of the world. It seems unlikely that the Humboldt Current contributes to the occurrence or distribution of ascidian fauna around the Galapagos islands.

Observations from the *Johnson-Sea-Link* have shown that ascidians are a very small part of the bathyal biomass which is mainly comprised of sponges, cnidarians and echinoderms. Other filter-feeding animals, such as bivalves or bryozoans, were also very seldom seen. Except for *Situla,* which is phytophagous and carnivorous, the other ascidians we report have the same diet as sponges: small suspended particles and dissolved substances. Sponges in Galapagos deep-water habitats are large, abundant, and diversified, but ascidians remain scarce even though they are relatively large.

## Acknowledgments

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